MUSCLE SPINDLES IN THE HUMAN FOETUS

by

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Introduction and historical review

When muscle spindles were first reported and subsequently described they were called „primitive bundles" and „muscle buds". The term „muscle spindles" was coined by KÜHNE (1863) on account of their shape. The majority of fibres within spindles (intrafusal fibres) resemble foetal muscle fibres in being of small diameter, having similar staining reactions and centrally-placed round *nuclei*. The multinucleate equatorial region was thought to indicate intense formative activity and therefore spindles were considered to be sites of successive reproduction of extrafusal muscle fibres by longitudinal division. (HASSAL, 1851; WEISMANN, 1861; KÖLLIKER, 1862 and KÜHNE, 1863). Although spindles had been found in healthy muscle of frogs, rats and mice, in man, FRAENKEL (1878) and MILLBACHER (1882) amongst others, regarded them as a pathological response to a wide variety of acute and chronic systemic diseases, infections, traumata and certain neurological disorders, including a radial nerve injury. BABINSKI (1886) who had originally seen them in a case of 'amyotrophy' following injury to the spinal cord, appears to have been the first to have examined muscle from a healthy individual. Having found spindles in this case, he came to the conclusion that not only were they normal structures, but also that they might be autonomous, functioning independently of extrafusal fibres. This hypothesis was based on the fact that the spindles were bounded by a connective tissue capsule with separate but communicating compartments for blood vessels, nerves and muscle fibres. ROTH (1887) finally established the spindle as a normally occurring structure with further observations on human and other mammalian muscles.

Ill defined nerve endings were depicted by KÖLLIKER (1862) and KÜHNE (1863). BABINSKI (1886) and ROTH (1887) both reported on their presence, but KIRSCHNER (1888) was the first to demonstrate unequivocally the nervous connexions of spindles in man and several other mammalian species, and to postulate their sensory function. In 1898 RUFFINI extended his own earlier work and published the most beautiful drawings of the various types of spiral and plate-like endings in the equatorial, juxta-equatorial and polar regions of intrafusal fibres in the cat. Since the latter types of ending were unlike extrafusal motor-end-plates, he doubted whether they had a similar function. SHERRINGTON (1894) proved the sensory function of the annulospiral endings and showed that at least a third of the fibres are afferent in a so-called purely motor nerve. In a series of experiments in the cat, TOWER (1932) clarified, but did not completely solve, the problem of distribution of motor, sensory and autonomic nerves to the spindle.

Later work has shewn that there are two, possibly three, types of intrafusal muscle fibre. Intermediate and large diameter fibres have equatorial nuclear bags and there are nuclear chains in the smallest fibres. These types of fibre differ in staining reactions, fibrillar structure and probably in twitch characteristics. Histological and electrophysiological work in the cat indicates that in addition to an autonomic supply, there are two distinct types of both motor and sensory innervation. The details of the present position are summarised in the proceedings of the Symposium on Muscle Receptors (BARKER 1961).

Although the analysis of structure and function is incomplete, spindles are known to play an essential role in the control of striated muscle. It is therefore of interest to trace their development in the human *foetus* and to attempt to correlate this with general development and with the evolution of foetal movements.

In limb buds, skeletal and muscular elements are developed *in situ* and differentiate proximo-distally. The main structures of the upper limb are distinguishable by the end of the 6th week and of the lower limb at the 7th. The brachial *plexus* is formed and nerves grow into the arm during the second half of the fourth week. At the beginning of the fifth, the lumbo-sacral *plexus* is formed and nerves begin to penetrate the lower limb. As shewn by BARDEEN and LEWIS (1901), the rudiments of all the skeleton, muscles and nerves are developed during the first two months and the adult condition is attained by subsequent increase in size and complexity of organs and by relative shifting of parts. These authors also noted that differentiation of muscle followed immediately after a motor nerve entered the region, but pointed out that this not necessarily imply a causal relationship.

MAVRINSKAIA (1960) reported that primitive motor and sensory endings are identifiable in human foetal intercostal muscles at about 7½ to 8 weeks. She studied these muscles in 67 foetuses aged from 5—16 weeks, using BIELSCHOWSKY—GROS silver impregnation and PORTUGALOV's histochemical test for acid-phospho-mono-esterase activity, on frozen sections. (No mention was made of the fixative or any decalcifying fluid.) In her view, motor and sensory endings were laid down simultaneously.

On the other hand, HEWER (1935) using both RANSON's and de CASTRO's silver impregnation on a smaller series of formalin-fixed foetuses aged from 6 weeks to full term, stated that sensory endings were almost completely developed before motor endings. For example, muscle spindles were well differentiated by 20 weeks, and at birth motor endings were rudimentary excepting in the tongue, intercostals and diaphragm. The cephalo-caudal and proximo-distal gradients of differentiation noted by BARDEEN & LEWIS (1901) applied to nerve endings, as well as to general tissue differentiation within the trunk and limbs. Apart from stating that an argentophilic *plexus* was present in all muscles by the eighth week, few details were given of the early formation of endings.

The most detailed and lavishly illustrated account of development of spindles and motor-end-plates in man has been given by CUAJUNCO (1940 & 1942). The observations on spindle and motor endings were confined to *biceps brachii* and a „few forearm muscles" in 25 foetuses of ages ranging from 10—39 weeks. The first dotlike motor endings and a few simple networks of sensory fibres around muscle fibres, in no way distinguishable from the rest of the muscle, were seen at the 11th week. As stated by HEWER (1935), the spindle capsule was forming in the 12th week. CUAJUNCO noted a well defined lymph space in the 14th week and lamination of the capsule in the 15th. The intrafusal fibres were differentiated by the 12th week and three types were reported. However, the presence of large, medium and small fibres was attributed to a growth phenomenon. Nuclear bag and nuclear chain fibres were also observed.

The number of spindles in *biceps* had doubled or trebled between the 10th and 12th weeks and numerical increase continued up to, but not beyond the fifteenth week, the period at which proliferation of the muscle cell ceases. Isolated spindles were the commonest, but grouped and tandem spindles were also present.

From the 11th week, the pattern of innervation became more elaborate and nerve fibres entered different parts of the spindle. Equatorial and juxta-equatorial nerve fibres branched and formed different types of spiral and „flower" endings. Others entered the polar regions and „typical" end-plates were formed. Non-myelinated fibres accompained vessels and the equatorial fibres began to myelinate at the fourteenth week. At this time all components of the adult spindle were present and recognizable, although the completely mature form was not achieved until the 24th—31st week. The latter point agrees with TELLO (1917), who was the first to investigate the development of innervation of the muscle spindle. His single human specimen was from a 6 month *foetus* and the illustration depicted the typical mature pattern of motor, sensory and autonomic nerve fibres. Once the mature form was achieved, there was no further change, other than an increase in the size of the spindle with the age of the *foetus*. This increase in size with age had been noted earlier by FORSTER (1902), and it led her to conclude that the spindle was „physiologically important and not an embryonic remnant."

The precise relationship between the onset of functional activity and the pattern of intramuscular innervation has been studied in a series of sheep foetuses (BARCROFT and BARRON, 1936 and 1937 and DICKSON, 1940). Unfortunately, no strictly correlated study is available for the human, but observations on spontaneous and reflex movements have been published.

Spontaneous intrauterine contractions are detectable at laparotomy at the 14th week but these are not detectable by the mother until the 16th or 17th week (WINDLE 1944). In highly artifical conditions after hysterotomy, reflex movements of head and trunk were elicited at about $7^1/_2$ weeks and of hand and foot at the 10th and 11th weeks respectively by HUMPHREY and HOOKER (1959). HOGG (1951) obtained reflex movements of head, hand and foot at $8^1/_2$, 11 and 12 weeks. In his series he examined the state of sensory nerves and associated structures of the skin of 19 foetuses of 8 to 14 weeks of menstrual age. Like HEWER (1935), he concluded that the adult form of the nerve endings was not a valid criterion of functional activity. From CUAJUNCO's histological findings, the simplest type of motor and sensory endings would appear to form at, or slinghtly after the onset of reflex contractions of the upper limb. The establishment of reflexes suggests that peripheral and central connections must exist and there is therefore need to re-examine the problem.

A full correlation of structure and function of developing muscle should include not only an account of the muscle fibres, but also of peripheral and central connexions of the motor and sensory nerves and tracts, together with observations upon peripheral and central myelination. In the present report an attempt will be made to report on the developing muscle spindles.

Material and Methods

Seven foetuses were used, their ages ranged from 57 days to 36 weeks of gestation. The three youngest were fixed in 7,5% formol-saline and the remainder in 10% formol saline. Details of each foetus and the material studied are set out in Table I. Several factors governed the choice of individual muscles. The spindle population is known to be high in small muscles of the hand and neck. (COOPER and DANIEL, 1956; VOSS, 1958); foetal intercostal, arm, forearm and hand muscles have been studied by other workers; the cephalo-caudal and proximo-distal gradient of differentiation could be investigated by taking the whole limb or a muscle from the neck and foot of a single individual and lastly, the small size of transverse sections of the neck makes it possible to study the whole *pharynx*, *larynx* and infrahyoid region. Although spindles have been reported in the two latter groups of muscles by LUCAS KEENE (1961) and VOSS (1958) respectively, there is still some lack of agreement on the subject. There appears to be no report of spindles in the pharyngeal muscles.

Specimens containing skeletal elements were decalcified in BENSLEY's fluid (50% Formic acid and 20% Sodium citrate in equal parts). Paraffin sections were cut at 10 μ and every section was mounted, all slides were stained, using Van GIESON's stain with iron haematoxylin and GLEE's silver (1946) impregnation on alternate slides. Findings will be reported in order from the oldest and most differentiated *foetus* to the youngest and least differentiated.

Observations

36 WEEK FOETUS. RB 123. FIRST DORSAL INTEROSSEUS MUSCLE OF HAND

Extrafusal muscle fibres were clearly cross-striated and the subsarcolemmal *nuclei* were rather rounder than in adult muscle and there was conspicuous difference in diameter between these and the intrafusal fibres (Fig. 1). There were numerous spindles, these appeared relatively much larger than in adult muscle.

The capsules and equatorial lymphatic space were conspicuous (Fig. 1 & 2). The intrafusal fibres were faintly cross-striated and stained rather more darkly than extrafusal fibres with VAN GIESON's stain. Nuclear chains and nuclear bags were identifiable (Fig. 1). Unfortunately, no completely longitudinal section of a spindle was found in the GLEE's preparations, but Figs. 3 & 4 taken from adjacent sections, show the entry of the large diameter sensory fibre and a few spiral turns in the equatorial region, and in Fig. 5 a medium sized fibre is seen approaching the juxta-equatorial region. Tendon organs and Pacinian corpuscles were well defined (Figs. 6 & 7), but motor nerve endings were small and simple (Figs. 8 & 9).

25 WEEK FOETUS. S 6/62. FIRST DORSAL INTEROSSEUS MUSCLE OF HAND

Crossstriation was well defined, the sub-sarcolemmal *nuclei* were large with conspicuous *nucleoli*. Spindles were also conspicuous, as the nuclear chains and bags stanied heavily and there was a well defined sub-capsular lymphatic space (Figs. 10 & 11). A motor nerve fibre destined for the two polar regions of one spindle is seen dividing the nerve trunk in Fig. 12 and simple extrafusal motor endings are shown in Figs. 13.

18 WEEKS FOETUS. S 25/62. 148 mm. C. R. LENGTH

A cephalo-caudal gradient of development and differentiation of muscle fibres and spindles was clearly demonstrated in this foetus, (Figs. 14 & 15 *rectus capitis posterior major* and Figs. 16 & 17 of *abductor hallucis* and the transverse head of *adductor hallucis* respectively.), The innervation of spindles was well differentiated in the sub-occipital muscle (Figs. 18, 19 & 20), but only fine and poorly stained fibres were found in the intrinsic muscles of the foot. However, the latter has been treated with a decalcifying agent, and this might account for some differences in staining of the nerve terminals, although general morphological characters would not be affected. In *rectus capitis posterior major* which measured 7 mm. in length, a few large diameter fibres had been broken at each end and dislocated from their surroundings by the sectioning. Two are shewn in Fig. 21, the longer measured 4.19 mm. suggesting that it might have extended throughout the muscle. Fibrous capsules had evidently been present in certain segments, for fragments were found at intervals, the proximal torn end of one on the longer fibre is seen in Fig. 22. Five separate fragments were present on this fibre and there were two on the shorter one.

There was an elaborate nerve supply to each of these fibres; and this was provided by large and finer diameter fibres approaching at intervals along the length of the muscle fibres in which no cross-striation was seen.

Both spiral and plate-like endings were present (Figs. 23 & 24). There were two main zones of innervation in the longer fibre and four in the shorter. Fibres of this type with similar innervation were found in several other sections of this muscle, and in an intercostal muscle of S 104/62, a large fibre with rudimentary zonal nerve endings was also found.

As expected, the density of innervation was high in all laryngeal muscles, spindles were identified in each and examples are shown from the *cricoarytenoideus posterior* and cricothyroid muscles, (Figs. 25, 26, 27 and 28).

In all muscles examined in this *foetus* (148 mm. C. R.), the spindles appeared relatively much longer than in the adult, for example, the intercostal spindle in Fig. 29 was 0.9 mm in length, and even then it was not certain that the ends were included in the section (see also Fig. 16). Nuclear chains and bags were identified in the intrafusal fibres in all muscles, although only nuclear chain fibres appear to be present in the one shown in Fig. 29. The capsules of spindles were defined in the laryngeal and neck muscles, but the lymphatic space was inconspicuous in the intercostal and foot muscles. In transverse sections of laryngeal and infrahyoid muscles it was very difficult to identify spindles unless the equatorial region was in the plane of section, for extrafusal *fasciculi* were small and there was no apparent difference in the diameters of the extrafusal fibres and the polar regions of intrafusal fibres. There was also no distinction in the size of intra-and extrafusal fibres in the foot, except in the equatorial region.

14 1/2 WEEK FOETUS. S 23/62. 106 mm. C. R. LENGTH

Cross-striation was visible in the muscles examined. The density of innervation of the laryngeal muscles was heavy. Spindles were present in these muscles and in the infrahyoid group. The nerve fibres were slender and finely branched, they lay in loose strands in the supporting tissue of the nerve trunks. In the longitudinal sections of sternomastoid there were many long slender spindles with nuclear bag fibres and the capsules were present (Fig. 30). Nerve fibres stained well in the nerve trunks and spiral endings were present in the equatorial region of the spindles, and in Fig. 31, part of this is visible together with a fine fibre running along the polar region. No motor endings were identified in the spindles. Simple dot-like endings were found on extrafusal fibres and were presumed to be motor endings.

13 WEEK FOETUS. S 16/62. 85 mm. C. R. LENGTH

In the intercostal muscles the extrafusal fibres were faintly but clearly cross-striated, *nuclei* were elongated, most of them were subsarcolemmal although some still appeared to be centrally placed. Fibres in the nerve trunks stained clearly and well in the muscles examined (Fig. 32). Spindles were identified by the multinucleate equatorial area in VAN GIESON preparations. In the GLEES preparations swellings containing many *nuclei* were present along the course of some fibres (Fig. 33). The *nuclei* were flanked by darkly staining and apparently amorphous material that had no counterpart in the older foetal or adult spindles. This zone is reminiscent of the early embryonic myoblasts (HEWER 1935) and of myoblasts grown *in vitro* (KONIGSBERG 1963). In relation to this region there was clear evidence of a connective tissue capsule and a nerve fibre can also be identified in Fig. 33. These areas were therefore identified as nuclear.

9 1/2 WEEK FOETUS. S 104/62. 46 mm. C. R. LENGTH

Extrafusal fibres were slender, faintly cross striated and contained large elongated or slight oval *nuclei* with well defined *nucleoli*. Connective tissue was present, but stained diffusely. The *rectus capitis posterior major* and the intercostal muscles contained numerous spindles, readily identified in the VAN

GIESON preparations by the multinucleate equatorial regions (Fig. 34). The capsule was present but thin and closely applied to the contained muscle fibres. Nerve fibres stained clearly, but were fine and no nerve endings were found in relation to the equatorial regions of spindles. A large and long unstriated and encapsulated fibre was found in an intercostal muscle, it had been dislodged and pulled across the section. A few darkly stained dots were found at intervals along the fibre, no spiralling fibres were seen and it is impossible to state whether these were early stages of plate endings found in S 25/62 (Figs. 23 & 24).

The muscles of the lower arm, forearm and hand were less well differentiated than the suboccipital muscle. In the hand the fibres were densely packed together and cross striation was extremely faint, especially in the silver preparations. In all three zones of the upper limb there were multinucleated fibres, clearly seen in the VAN GIESON stained sections (Fig. 35). Nerve fibres were present in arm and forearm and stained poorly, none was found in the hand. In the forearm nerve fibres ran along and across muscle fibres. No endings were identified in relation to the presumptive equatorial region found in an unidentified muscle of the forearm (Fig. 36).

57 DAY (8 WEEKS, 1 DAY) FOETUS. R 37. 33 mm. C. R.

The pre and post vertebral muscles of the neck and developing *larynx* contained numerous bundles of nerve fibres, some formed plexuses, but it was not possible to determine whether these were forming any specific type of sensory or motor ending. The muscle fibres were of small diameter. No spindles were identified in the neck.

In the intercostal muscles, fine rather poorly stained nerve fibres were seen to run between muscle fibres and to make a few loops around them. These muscle fibres had no other distinguishing features. A few muscle fibres cut in cross section were seen to have peripherally situated myofibrils. Two of these fibres were seen to be encircled by nerve fibres (Figs. 37 & 38).

In the arm and forearm there was no clear evidence of cross striation of muscle fibres. Fine nerve fibres ran in loosely knit strands in nerve trunks which were disproportionately large on comparison with the muscle masses. In the arm muscles there were a few grouped fibres with central, multinucleated zones. These structures were confined to the lower third of *biceps brachii, brachialis* and *triceps* and a few were seen in the forearm. One of these multinucleate structures, seen at successively lower focal planes (Figs. 39, 40 & 41) can be seen to have a delicate connective tissue capsule, a nerve fibre which is breaking up in relation to the multinucleate region that forms a localized swelling in a slender fibre. On this evidence it seems reasonable to suggest that this structure is a muscle spindle.

Discussion

In her study of intercostal muscles, MAVRINSKAIA (1960) has provided the earliest histological and histochemical evidence of neuromuscular contacts in man. She reported the simultaneous formation of simple motor and sensory endings between 7 ½ to 8 weeks. The youngest *foetus* in the present investigation was 57 days old and findings would support her claim that neuromuscular contact was made in certain muscles by this time. *Hewer* (1935) found an argentophilic plexus of nerves in all muscles at 8 weeks but gave no details of the early stages of formation of endings. In her view differentiation of sensory endings was in advance of motor ones, for spindles were well defined by 20 weeks and, with few exceptions motor endings were rudimentary at birth. CUAJUNCO (1940 & 1942) found no sign of neuromuscular contact in *biceps brachii* before the 11th week, and since well authenticated movements had been reported in younger foetuses, he suggested that myogenic contraction due to chemical stimulation might occur in the absence of effective innervation. Myogenic contraction of skeletal muscle has been reported in selachian embryos, but there is no evidence that it occurs in mammalian ones (BARRON, 1941). Furthermore, it would seem unnecessary to invoke such an explanation because rudimentary central connexions of neurones of spiral reflex arcs are demonstrable shortly before the time at which the contractions are elicited (BARRON 1941) and the work of MAVRINSKAIA (1960) and the present study both provide evidence suggesting that peripheral contacts also are formed in advance of the onset of reflex movements (cf HOGG 1941; HUMPHREY & HOOKER, 1959).

Some of the discrepancies between the various reports might be due to difficulties in accurately estimating foetal ages; individual variations between foetuses of the same age (CUAJUNCO, 1940) and variations in histological techniques. As CUAJUNCO's material came from the CARNEGIE collection, the estimate of age was certainly of the highest order of accuracy. Individual variation between foetuses would be unlikely to account for at least three week's difference in the formation of nerve endings, especially when general morphological features of the muscle fibres corresponded so well with these reported by others for foetuses of similar age. Frozen sections and histochemical techniques were used by MAVRINSKAIA alone, the silver techniques varied for each investigation. RANSON's and de CASTRO's impregnations were used by HEWER, AGDUHR's modification of BIELSCHOWKY's by CUAJUNCO, BIELSCHOWSKY GROS by MAVRINSKAIA, a GLEES method for paraffin sections was used in the present study. Previous authors did not mention decalcifying fluids, but must have employed them when skeletal elements were present, BENSLEY's fluid was used here. Variations in the time and duration of fixation are probably highly significant since FITZGERALD (1960) has shewn that this is critical for revealing fine axonal terminals. CUAJUNCO (1940 & 1942) used 20 % formalin, there was no specific mention of fixative by MAVRINSKAIA, and in this study 7,5 % and 10 % formol-saline were employed. The strong fixative might well have influenced the staining of the nerve endings in CUAJUNCO's series, although the definition of other structures was outstandingly clear.

The structure of the equatorial region of the spindle in the 57 day foetus (Figs. 39, 40 & 41) raises problems. It resembles the myotubes described by many, including HEWER (1935) and MAVRINSKAIA (1960). These are considered to give rise to muscle fibres and they have been in tissue culture in the absence of nerve fibres.

The first evidence of sensory innervation is the formation of spiral turns of nerves around muscle fibres, in *no way distinguishable* from their neighbours. If, as evidence suggests, the structure shewn in (Figs. 39, 40 & 41) is a spindle, there must have been a dedifferentiation towards the more primitive type of

Table I.

SERIAL NO.	C. R. LENGTH (mm)	ESTIMATED AGE (weeks)	MATERIAL SECTIONED	PLANE OF SECTION
RB 123		36	First dorsal interosseus muscle (hand) left right	L. S. T. S.
S. 6/62	225	25	First dorsal interosseus muscle (hand)	T. S.
S 25/62	148	18	Rectus capitis posterior major left right Neck Intercostal muscles and ribs Foot (left)	L. S. T. S. T. S. In long axis of ribs L. S. in plane of sole
S 23/62	106	$14^1/_2$	Tongue Block of larynx, trachea, infrahyoid muscles, pharynx & oesophagus Sternomastoid (left)	T. S. T. S. L. S.
S 16/62	85	13	Intercostal muscles & ribs Hand (left)	In long, axis of ribs T. S.
S 104/62	46	$9^1/_2$	Rectus capitis posterior major (left) Intercostal muscles & ribs Forearm and hand	L. S. In long axis of ribs L. S.
R 37	33	8 weeks 1 day	Neck Intercostal muscles & ribs Upper limb (left)	T. S. In long axis of ribs L. S.

structure in relation to the sensory fibre. The other alternative would be to interpret the myotube as the anlarge of the spindle, and for this there is no support.

Although attention has been directed chiefly to developing spindles, this investigation would support HEWER's statement that differentiation of motor-endings lags behind that of the sensory endings. The motor nerve endings in the hand at 36 weeks were simple and lacked elaborate end-arborisations. However, this may be a reflexion of the small size of extrafusal muscle fibres and not evidence of a lag in development. In the chick SHEHATA (1961) showed that as the diameter of the muscle fibre increased with age, the motor-end-plate increased in size and nerve terminals became more elaborately branched. It was suggested that this increase in branching was a normal form of terminal sprouting produced by minute detachments of nerve terminals due to the enlargement of the muscle fibres beneath them. The process of terminal sprouting of normal nerve fibres has been seen in partial denervation of animal and human skeletal muscle (HOFFMAN, 1951; and COËRS & WOOLF, 1959).

The long, encapsulated muscle fibres of large diameter found in the sub-occipital muscle of S 25/62 and in the intercostal muscle of R 37 are of interest. The well defined pattern of innervation in the older *foetus* resembles that found previously on relatively short segments of muscle in human extrinsic eye muscles (DOGIEL, 1906; DANIEL, 1946); facial muscles (KADANOFF, 1956) and laryngeal muscles (LUCAS KEENE, 1961); and in facial muscles of rabbit and rat by BOWDEN & MAHRAN (1956). ALDERSON (1962) also saw them in large numbers in intercostal muscles of the cat. These muscle fibres do not resemble the large diameter intrafusal fibres, because no evidence of a nuclear bag was found along their length. One end of one of the fibres was longitudinally striated, and the possibility of this being a very long and slender spindle with poorly stained intrafusal fibres must be considered. However the strikingly different pattern of innervation and absence of any sign of a dilated equatorial region, render this possibility unlikely.

Constant or nearly constant activity is a feature shared by all the muscle groups in which these spiral endings have been reported and in addition, precise and delicate movements are executed by most, if not all. The movements of respiration are graduated precisely and the suboccipital muscles might be considered to supply the fine adjustment for the position of the head. These specialised nerve fibres might represent some specialised form of low threshold stretch receptors.

This investigation confirm previous evidence of a cephalocaudal gradient of differentiation of muscle and nerve fibres and endings (see S 25/62). The fact that no definite endings were found in the transverse sections of the neck of R. 37., whilst they were seen in distal parts is almost certainly due to the fact that sections in this plane are not as suitable as longitudinal sections for detecting spindles in foetuses younger than three months. The intra and extra-fusal fibres are of the same diameter except in the equatorial region (Figs. 39, 40 & 41), and although there was evidence of a capsule, the subcapsular space was not defined. In longitudinal sections the spindles are conspicuous, and are apparently larger and more closely packed than in the adult. This relatively large size in the *foetus* may be attributable to the fact that after the 15th week there is no numerical increase in the spindle population (CUAJUNCO, 1940).

The presence of spindles in all laryngeal and infrahyoid muscles has been confirmed, but so far there is no evidence of spindles in the constrictors of the pharynx.

There is clear evidence to support HEWER (1935) and HOGG (1941) in their statement that the adult or mature form of nerve endings is no valid criterion of functional capacity. This view is supported by collateral evidence of restoration of function in the presence of abnormal endings after regeneration of injured peripheral nerves in experimental animals and man (GUTMANN & YOUNG, 1944; BOWDEN & GUTMANN, 1944). Whilst rudimentary spindles, motor endings and central connexions of their nerves appear to be established before movements occur, it is evident that further differentiation of spindles is not dependent upon active movement of the *foetus*. Well differentiated and apparently healthy spindles (Fig. 42) were found in the gluteal muscles of a child of 2 years with congenitally fixed hips due to *arthrogryphosis multiplex*.

Clearly further investigations using histochemical techniques, FITZGERALD's fixative for demonstrating fine nerve terminale and a variety of silver stains is required. Coupled with this electronmicroscopy is essential in order to find the earliest evidence of neuromuscular contacts and peripheral myelinogenesis

Summary

1. The development of neuromuscular spindles in the neck, laryngeal, infrahyoid, intercostal and limb muscles has been studied from seven human foetuses of ages ranging from 57 days to 36 weeks.

2. Evidence suggests that neuromuscular spindles are formed before the onset of reflex movements of the limbs and trunk of the *foetus*.

3. Large diameter muscle fibres with spiral and plate-like endings have been found in suboccipital and intercostal muscles. These resemble those reported by DOGIEL in extrinsic eye muscles.

4. The functional significance of the findings is discussed.

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Legends for figures

ALL SECTIONS ARE 10 μ THICK

Figs. 1—9 inclusive. lst dorsal interosseus muscle (hand). 36 week foetus RB. 123

Figs. 1 & 2 — Adjacent oblique sections of muscle spindle. (VAN GIESON and iron haematoxylin stain.)
a) capsule.
b) ~~lymphatic space~~.
c) intrafusal fibres.
d) nuclear bag fibre.
e) nuclear chain fibre.
f) nerve trunk.

Figs. 3 & 4 — Oblique sections of muscle spindle. 3 & 4 are adjacent to each other. (GLEES stain.)
a) *capsule.*
b) equatorial region of spindle.
c) nerve fibre approaching equatorial region.
d) part of annulo-spiral ending.
e) medium sized nerve fibre approaching juxtaequatorial region.
f) intrafusal muscle fibres.
g) nerve trunk.

Fig. 6 — Tendon organ (GLEES stain).

Fig. 7 — Pacinian corpuscle close to musculo-tendinous junction. (VAN GIESON and iron haematoxylin stain.)

Figs. 8 & 9 — *a)* motor-end-plates on extrafusal fibres.
b) intramuscular nerve trunk.

Figs. 10—13 inclusive. lst dorsal interosseus muscle (hand). 25 week foetus S6/62.

Figs. 10 & 11	L. S. of equatorial regions of two spindles. (VAN GIESON and iron haematoxylin stain.) *a)* extrafusal muscle fibres. *b)* poles of intrafusal fibres. *c)* nuclear bag fibre. *d)* nuclear chain fibre.
Figs. 12	Tangential section of equatorial region of spindle. (GLEES stain.) *a)* laminated capsule of spindle. *b)* equatorial region. *c)* lymphatic space. *d)* dividing motor nerve fibre in intramuscular nerve trunk. *e)* juxta-equatorial motor endings.
Fig. 13	Extrafusal end plates. (GLEES stain.)

Sections 14—29 inclusive. 18 week foetus. S 25/62. 148 mm C. R. length.

Figs. 14 & 15	L. S. *Rectus capitis posterior major.* (VAN GIESON and iron haematoxylin.) Note high density of spindle population in Fig. 14.
Figs. 16 & 17	L. S. *Abductor hallucis* and transverse head of *adductor hallucis* respectively. for figs. 14—17 inclusive note: *a)* equatorial region of spindle. *b)* poles of intrafusal fibres. *c)* capsule. *d)* extrafusal fibres.
Figs. 18, 19 & 20	L. S. *Rectus capitis posterior major.* (GLEES stain.) *a)* equatorial region of spindle. *b)* polar region. *c)* capsule. *d)* extrafusal muscle fibres. *e)* nerve supply of equatorial region. *f)* nerve to juxta equatorial region. *g)* motor fibre to intrafusal muscle.
Figs. 21—24	L. S. *Rectus capitis Posterior.* (GLEES stain.)
Fig. 21 Fig. 22	*a)* & *b)* two large diameter muscle fibres dislocated across main section. *c)* torn fragment of capsule on fibre of Fig. 21. *d)* extrafusal fibres (cf with fibre a)
Figs. 23 & 24	Details of innervation of large diameter fibre (of Fig. 21). P plate ending S spiral nerve fibres.
Figs. 25, 26 & 27	T. S. of *cricoarytenoideus posterior.*
Fig. 25	VAN GIESON & iron haematoxylin stain. Glees stain.
Figs. 26 & 27	GLEES stain. *a)* Equatorial region of spindle. *b)* capsule of spindle. *c)* extrafusal muscle *fasciculus.* *d)* nerve trunk. *e)* nerve fibres approaching equatorial regions of compound spindle.
Fig. 28	L. S. Cricothyroid muscle. GLEES stain. Note innervation of equatorial region.
Fig. 29	L. S. of spindle in intercostal muscle VAN GIESON and iron haematoxylin.

Fig. 30 and 31. L. S. Sternomastoid of 14½ week foetus. S23/62 106 mm C. R. length.

Fig. 30	VAN GIESON & iron haematoxylin stain.

Fig. 31 GLEES stain.
a = nuclear bag fibre
b = nuclear chain fibre
c = capsule
d = nerve trunk
e = spiral nerve ending
f = nerve fibre approaching polar region of intrafusal fibre.

Figs. 32 & 33. 13 week foetus. S16/62 85 mm. F. R. length

L. S. Intercostal muscle. (GLEES stain.)

Fig. 32 *a)* extrafusal fibre.
b) nerve fibres.
c) multinucleate presumptive equatorial region.

Fig. 33 *a) b) c)* as for Fig. 32.
d) capsule of spindle.

Figs. 34—36. 9½ week foetus. S104/62 46 mm C. R. length

Fig. 34 L. S. *Rectus capitis major posterior.* (VAN GIESON and iron haematoxylin stain.)
a) extrafusal fibres.
b) equatorial region of spindle.

Fig. 35 L. S. Interosseus muscle of hand. (VAN GIESON & iron haematoxylin.)
a) equatorial region of spindle.

Fig. 36. L. S. Muscle of forearm. (GLEES stain.)
a) presumptive equatorial region of spindle resembling myotube.

Figs. 37 to 41. 57 day foetus. R 37. 33 mm. C. R. length

Figs. 37 & 38 Intercostal muscle. (GLEES stain.)
a) T. S. of muscle fibres with peripherally placed myofibrils.
b) Nerve fibres looping round muscle, possibly early formation sensory ending.

Figs. 39, 40 & 41. L. S. *Biceps brachii.* (GLEES stain, successively lower focal planes.)
a) extrafusal fibres.
b) capsule.
c) nerve fibre & fine terminals.
d) details of multinucleate zone, presumptive equatorial region.
e) small diameter muscle fibre, presumptive polar regions.

Fig. 42. T. S. of spindle in *gluteus medius* of child of 2 years with *arthrogryphosis multiplex* and congenitally fixed hips. (ROMANES silver stain.)
a) capsule.
b) intrafusal fibres of large, medium & small diameter.

Fig. 1.

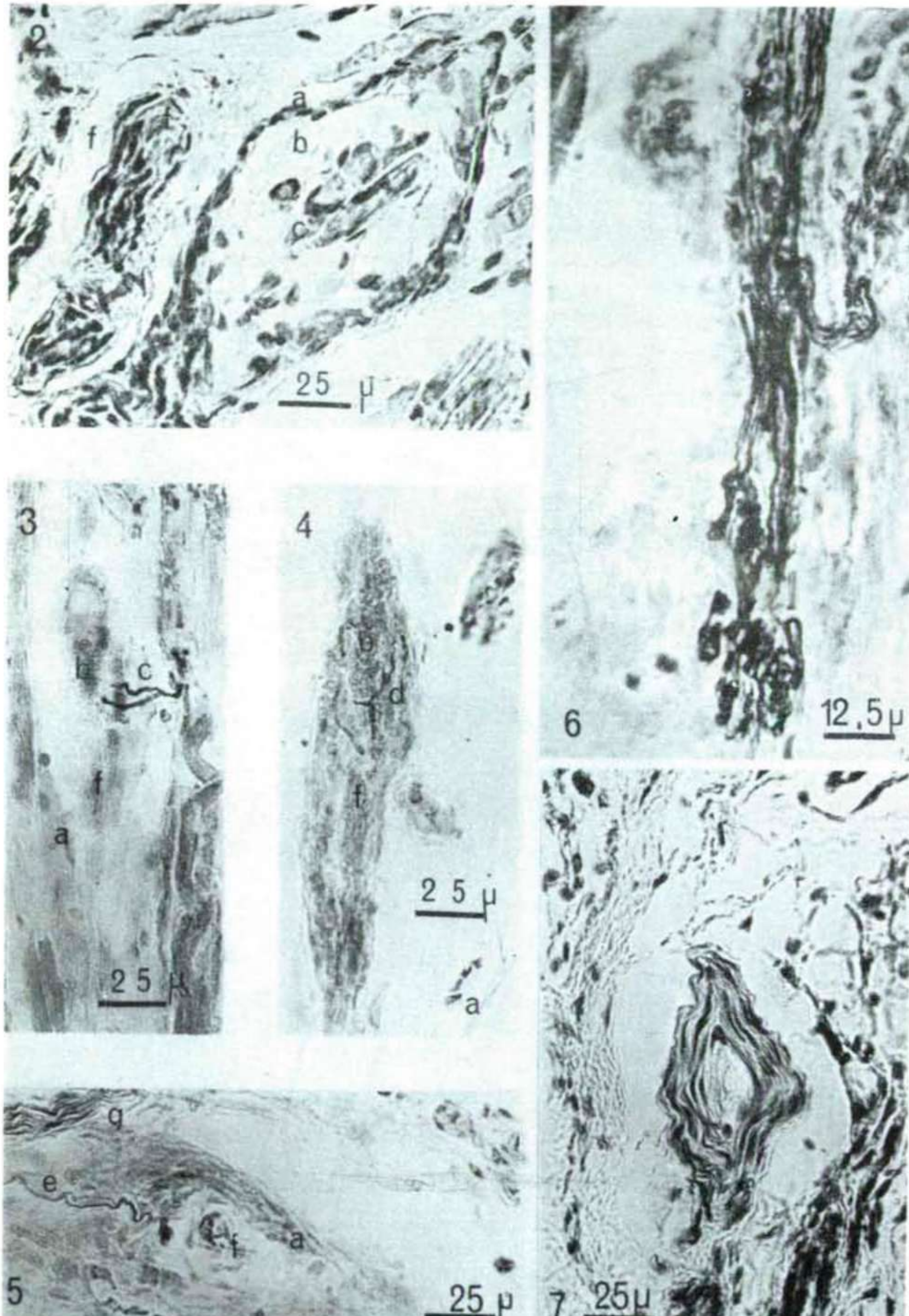
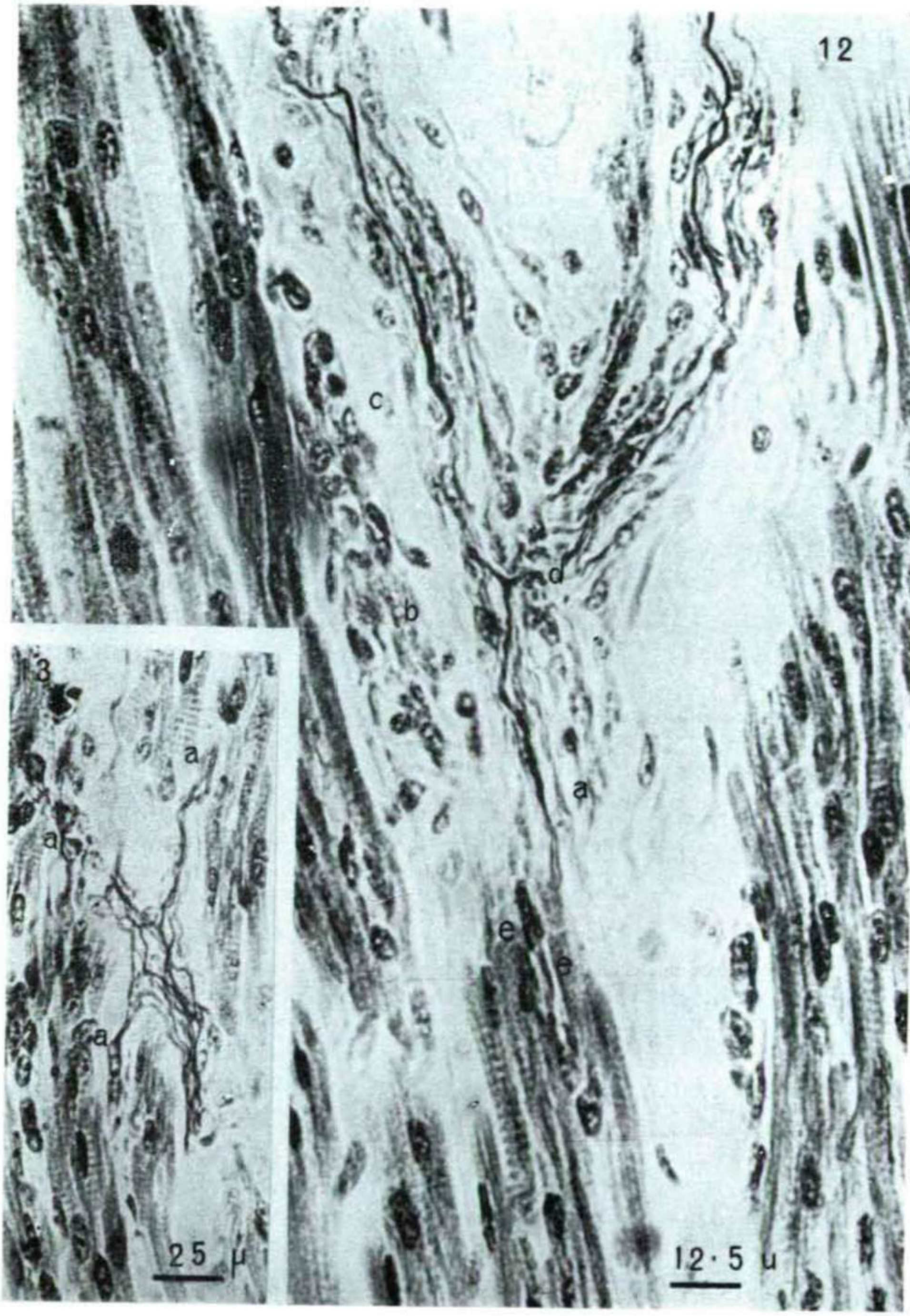


Fig. 2.

Fig. 3.



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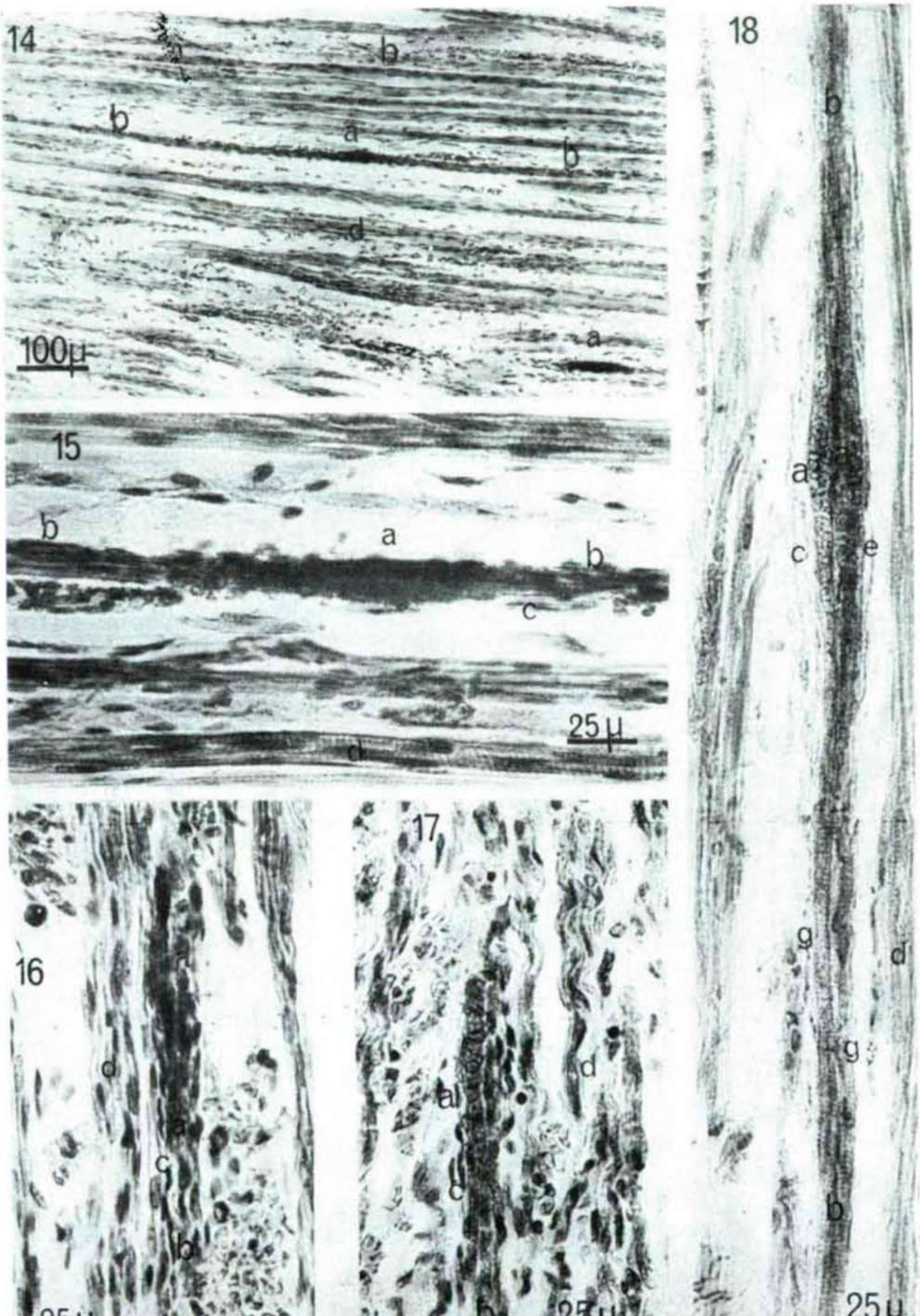
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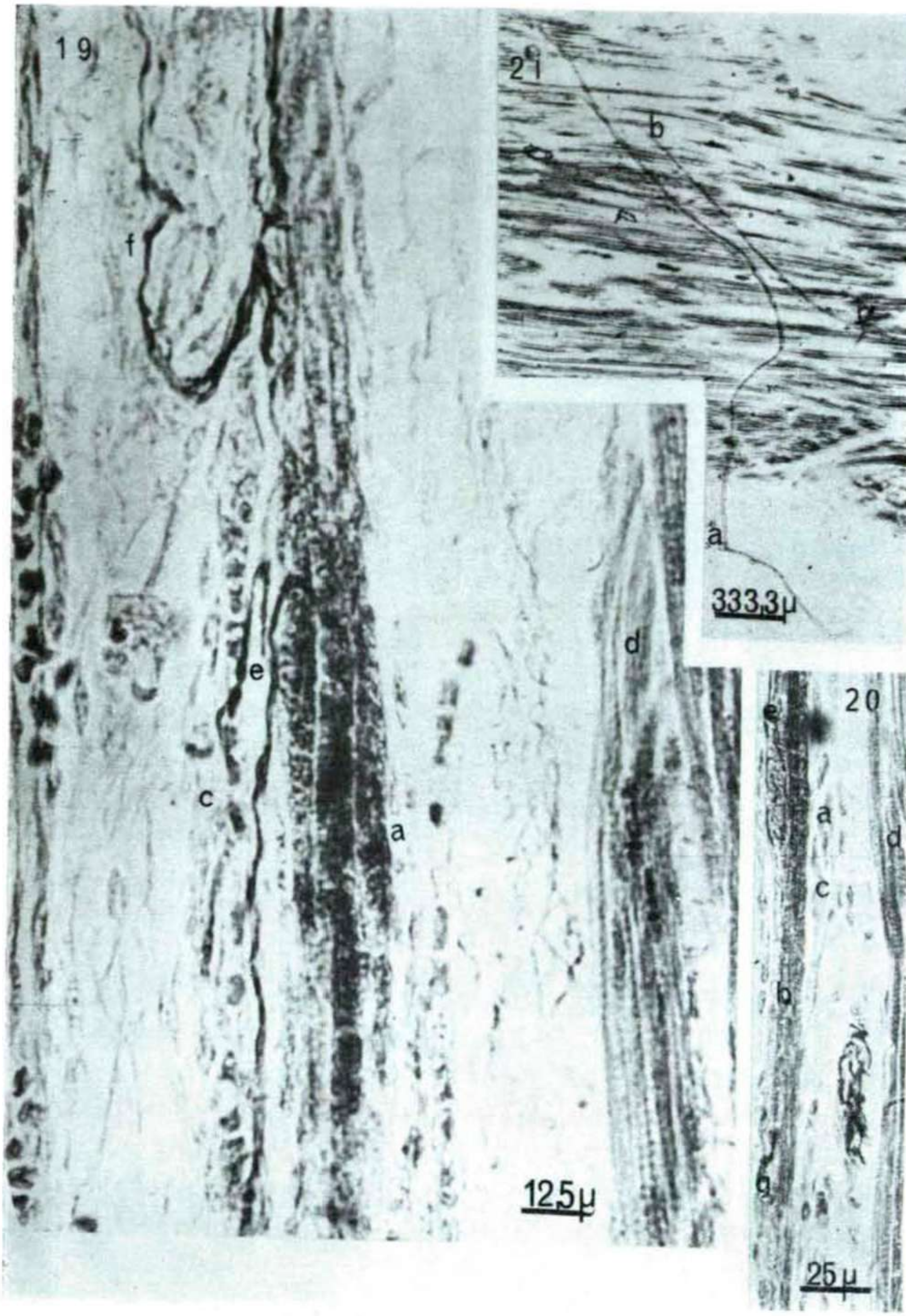
Fig. 5.



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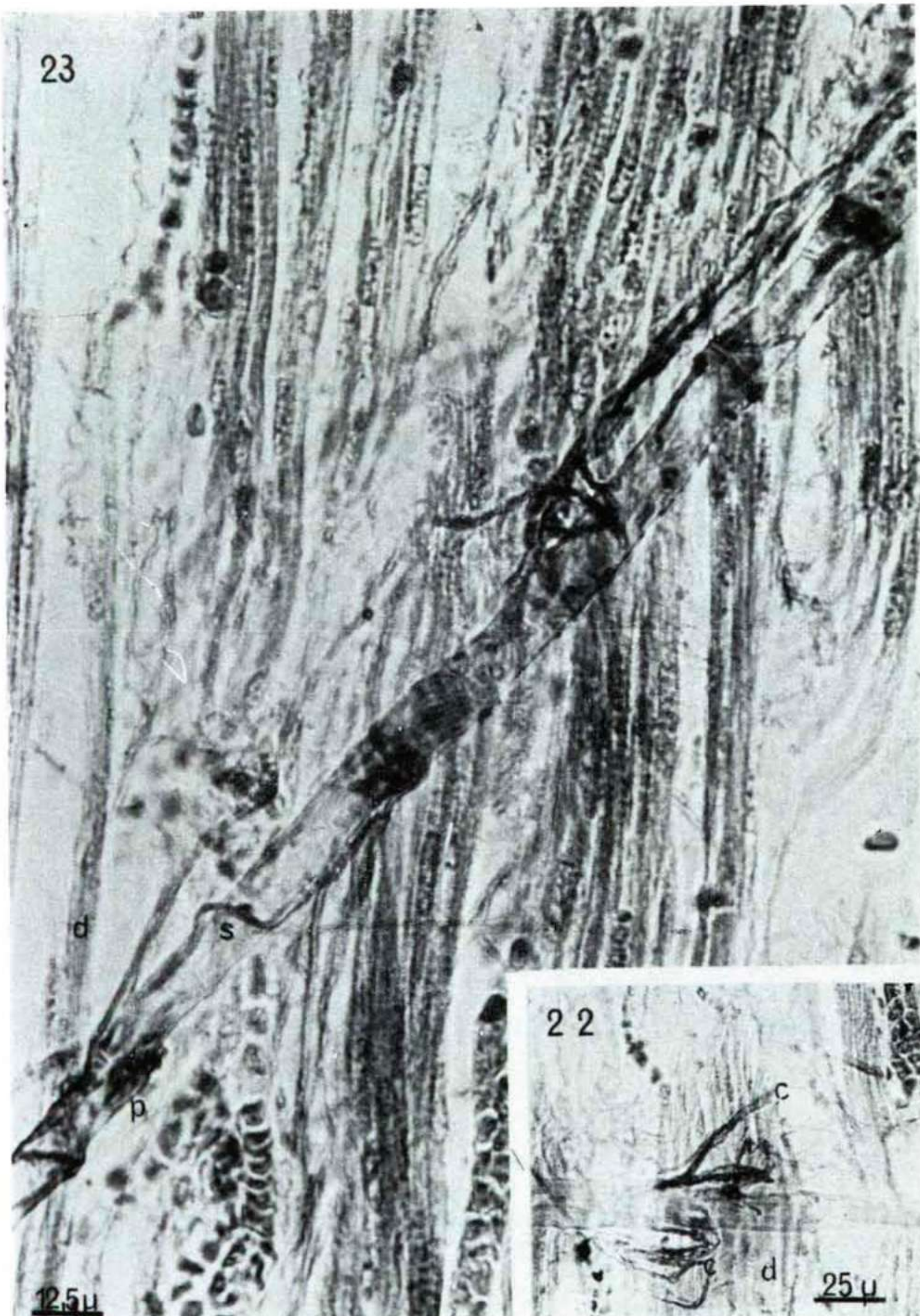

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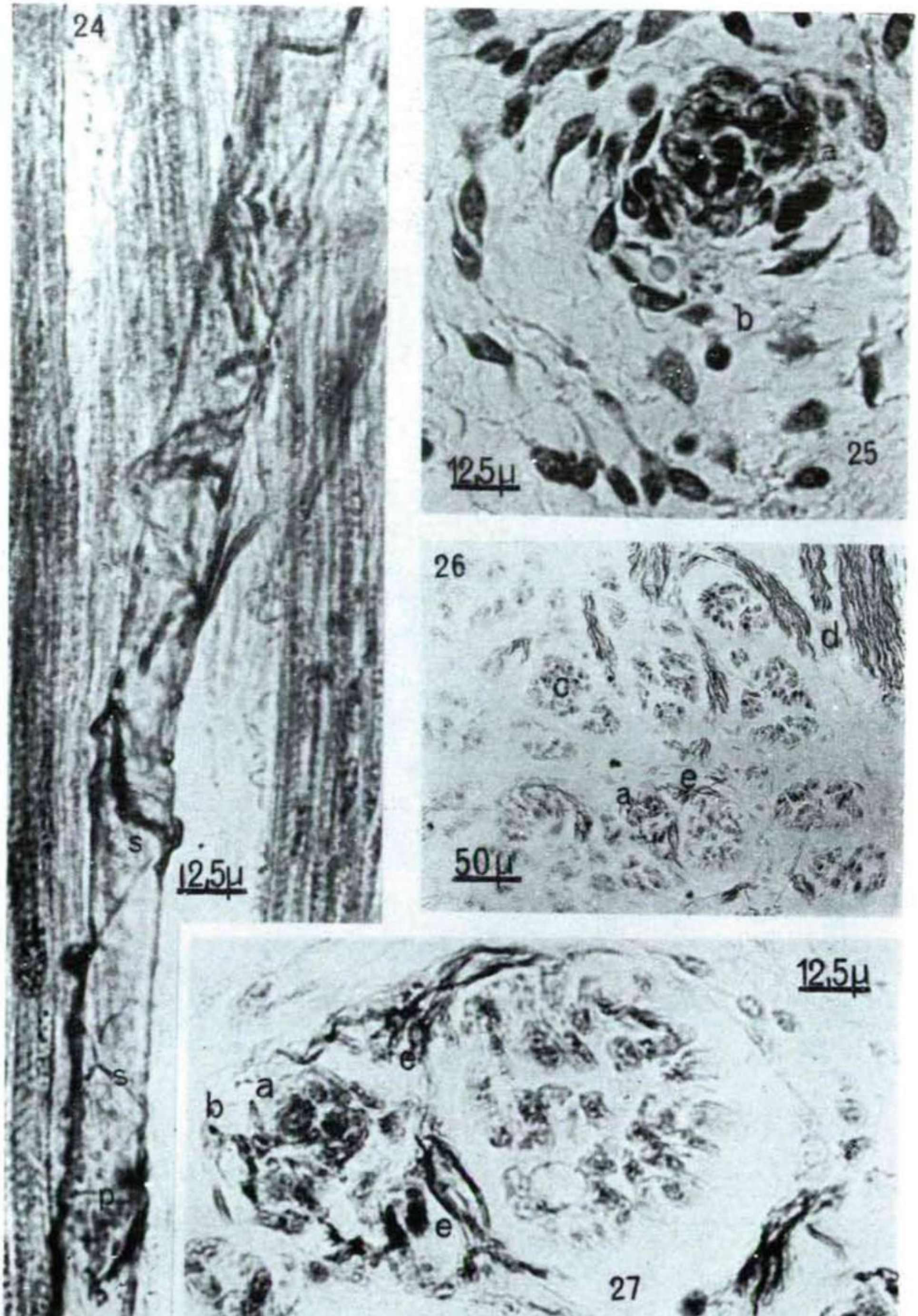

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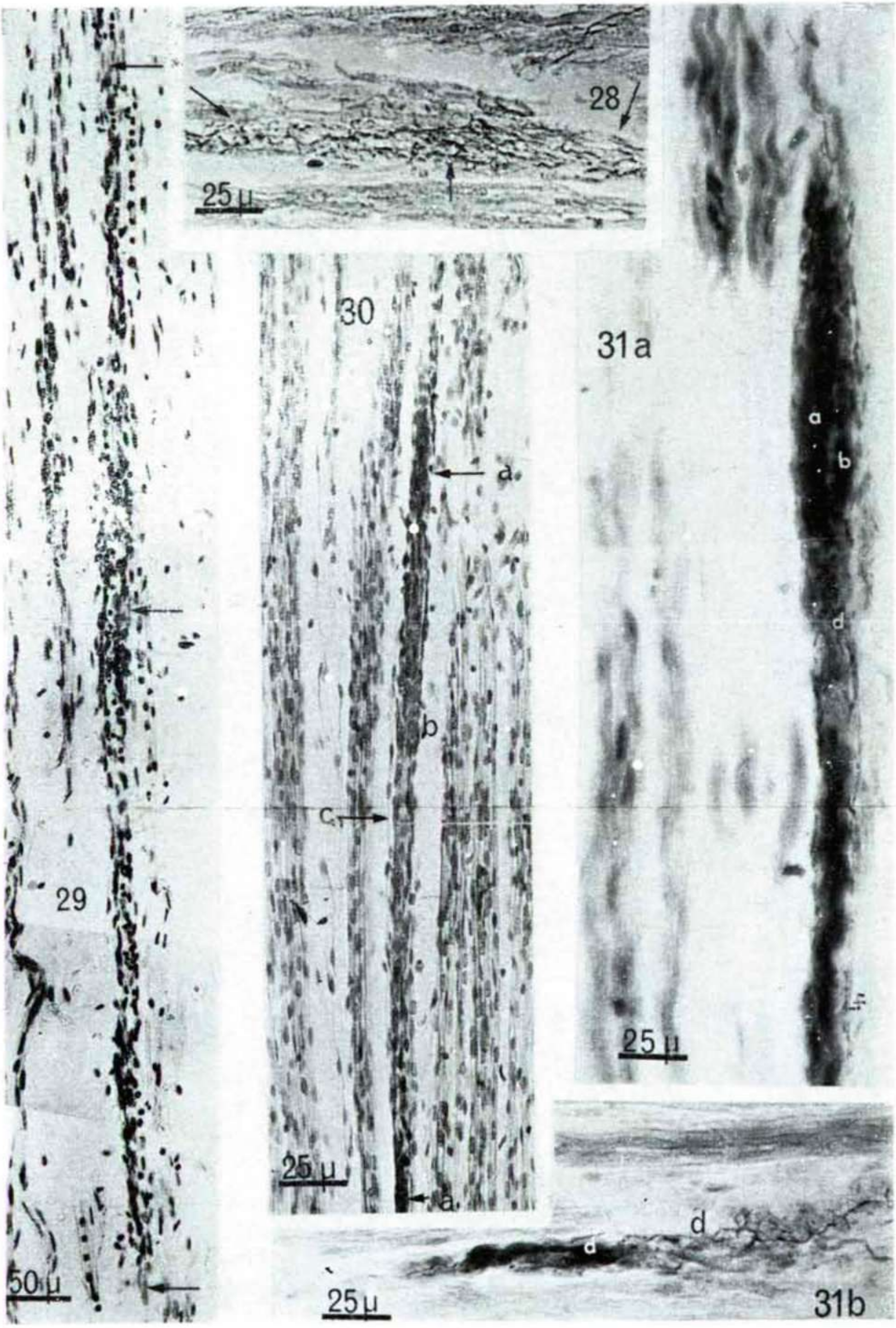

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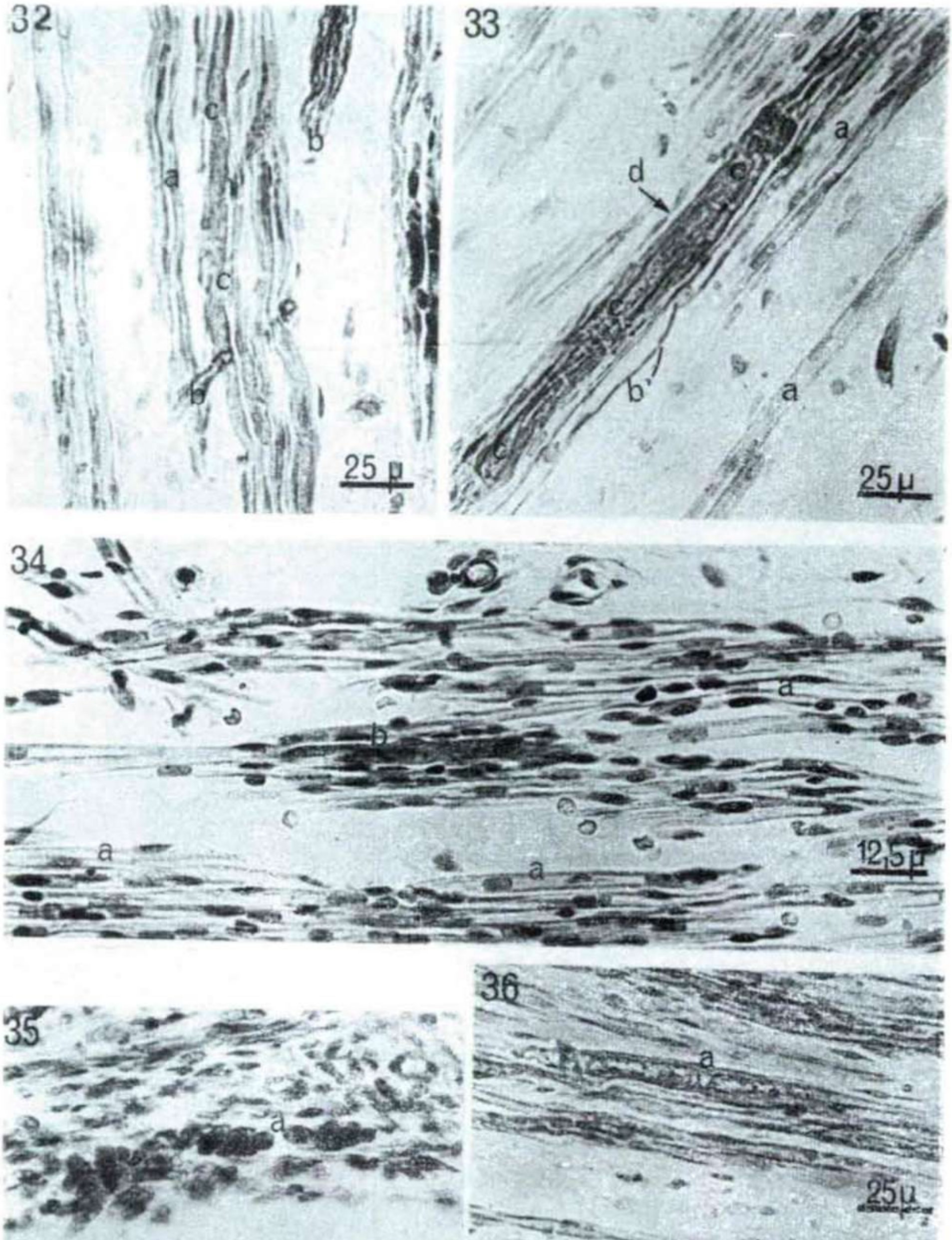

Fig. 10.

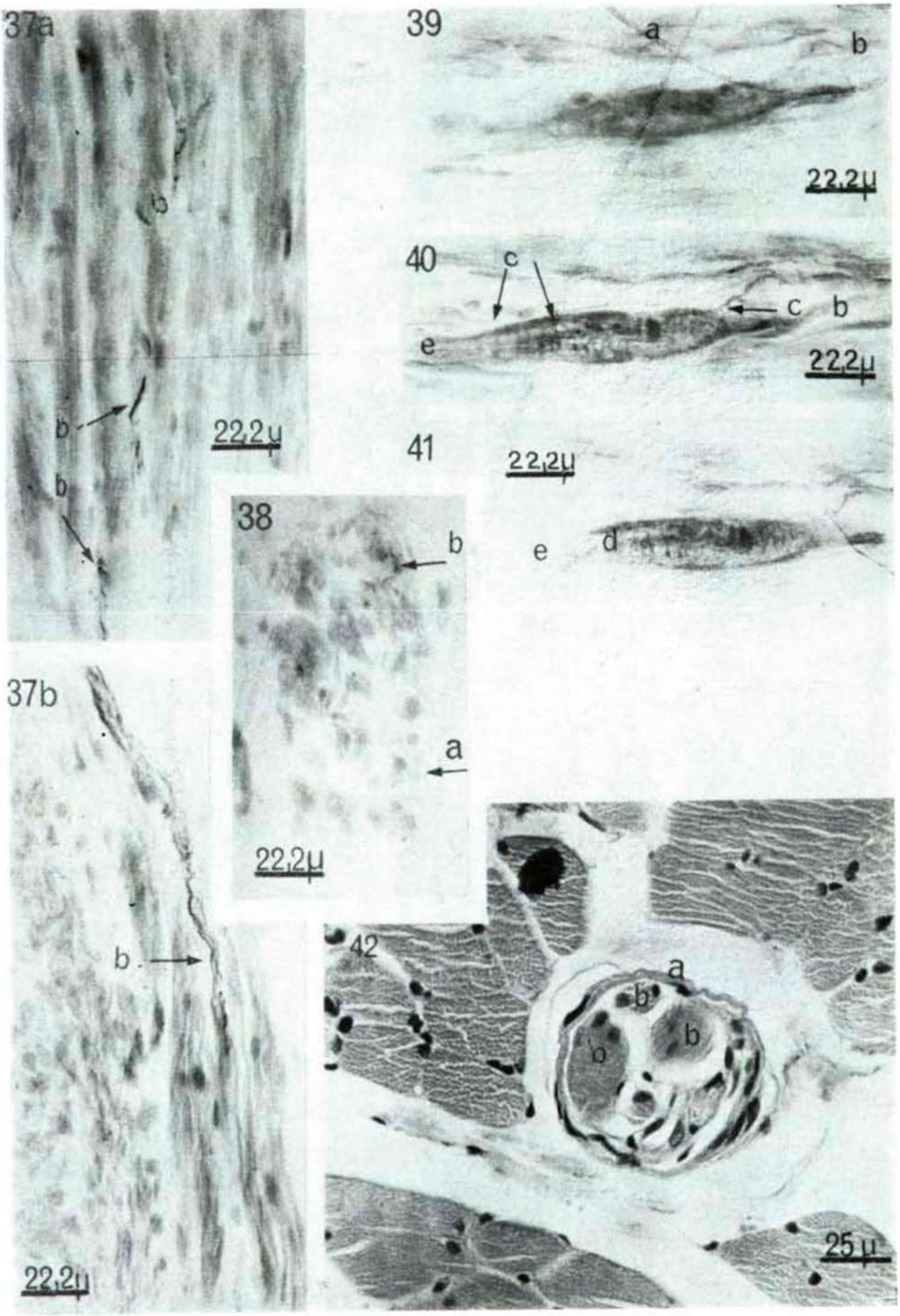
37a
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Fig. 11.